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Association of neutrophil-lymphocyte ratio with metabolic syndrome and its components in Asian Indians (CURES-143)

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ABSTRACT

Background: Metabolic syndrome (MS) is the state of chronic low grade inflammation. This study looks at the relationship of neutrophil-lymphocyte ratio (NLR) in subjects with and without MS in Asian Indians.
Methods: Study subjects (n = 754) were recruited from the Chennai Urban Rural Epidemiology Study. MS was defined using the National Cholesterol Education Program-Adult Treatment Panel (NCEP-ATP) III criteria modified for waist according to World Health Organization Asia Pacific guidelines. A complete hemogram was done in all subjects using a five-part hematology analyzer (model SF-3000; Sysmex, Kobe, Japan). The NLR was calculated as the ratio between counts for neutrophils and total lymphocytes in subjects with (n = 422) and without (n 332) MS and correlated with number of metabolic abnormalities in those with MS.
Results: Subjects with five metabolic abnormalities had the highest NLR, and with decreasing number of metabolic abnormalities, the NLR decreased linearly (p for trend <0.001). Logistic regression analysis revealed that even after adjusting for age, gender and body mass index, MS was strongly associated with NLR ($p < 0.001$).
Conclusion: Among Asian Indians, NLR is correlated with MS and also with the number of metabolic abnormalities.

1. Introduction

The term metabolic syndrome (MS) refers to a constellation of several cardiometabolic abnormalities such as abdominal obesity, insulin resistance, hyperglycemia, hypertension and dyslipidemia. Several reports have shown that MS increases the risk of diabetes by 2 fold and that of CVD by 5 fold (Grundy, Cleeman, Daniels, et al., 2005; Kaur, 2014). According to estimates by the International Diabetes Federation (IDF) almost one quarter of the world's adult population has MS (International Diabetes Federation, n.d.). The prevalence of MS is reported to be high in Asian Indians (Enas, Mohan, Deepa, et al., 2007). A report showed that 25.8% of the general population and 50% of subjects with type 2 diabetes have MS (Deepa, Farooq, Datta, et al., 2007).

Numerous studies have shown an association of MS and insulin resistance (IR) with inflammation. Two hypotheses have been proposed to explain the relationship of MS with inflammation. The first states that chronic low-grade inflammation leads to metabolic disturbances, which in turn leads to IR (Fernández-Real & Ricart, 2003). The second suggests that altered glucose and lipid metabolism trigger inflammation which results in IR (Shoelson, Lee, & Goldfine, 2006). The relationship of systemic inflammatory markers high-sensitivity CRP (hs-CRP), TNF-α and IL-6 with MS and IR has been shown in several studies (Emanuela, Grazia, de Marco, et al., 2012; Gokulakrishnan, Deepa, Sampathkumar, et al., 2009; Gokulakrishnan, Deepa, Sampathkumar, et al., 2009). Further, these systemic markers act as strong prognostic factors for the future onset of diabetes and CVD. However, most of these markers are time consuming and expensive. Recently it has been shown that there is a strong relationship between white blood cells (Gokulakrishnan, Deepa, Sampathkumar, et al., 2009) and more specifically the ratio of neutrophils and lymphocytes (NLR) and several metabolic diseases, like diabetes and CVD (Bhat, Teli, Rijal, et al., 2013; Shiny, Bibin, Shanthirani, et al., 2014). NLR is also known to be associated with systemic pro-inflammatory cytokines (Guthrie, Charles, Roxburgh, et al., 2013; Kantola, Klintrup, Väyrynen, et al., 2012; Motomura, Shirabe, Mano, et al., 2013).NLR has been shown to be a more valuable marker for CVD than WBC, as it is less likely to be influenced by physiological conditions (Bhat et al., 2013). Estimation of NLR is attractive as it is easy to perform, readily available even at remote locations and is inexpensive. There is one report in a Turkish population showing an

Conflict of interest: None.

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association of NLR with MS (Buyukkaya, Karakas, Karakas, et al., 2014). This study investigates the association of NLR with MS in Asian Indians, who are known to have an increased susceptibility to type 2 diabetes and premature coronary artery disease (Mohan, Sandeep, Deepa, et al., 2007).

2. Methodology

The study subjects were recruited from the Chennai Urban Rural Epidemiological Study (CURES), an epidemiological study conducted on a representative population (≥20 years old) of Chennai (formerly Madras), the fourth largest city in India. The methodology of the study has been published elsewhere (Deepa, Pradeepa, Rema, et al., 2003). In brief, 26,001 individuals were recruited for phase 1 of CURES, using a systematic random-sampling technique; subjects with self reported diabetes receiving treatment were classified as "known diabetes subjects." Fasting capillary blood glucose was determined using an OneTouch® Basic® glucometer (Lifescan, a Johnson & Johnson Company, Milpitas, CA) in all subjects. Details of the sampling are described in our website (www.drmohansdiabetes.com/bio/WORLD/pages/pages/ chennai.html). In phase 3 every 10th subject in phase 1 (n = 2600) was invited for clinical, biochemical, microvascular, and detailed eye examinations (Rema, Premkumar, Anitha, et al., 2005; Unnikrishnan, Rema, Pradeepa, et al., 2007). Phase 3 had a response rate of 90.4% (2350/2600 subjects participated). For the present study, every third subject from phase 3 of CURES (n = 783) maintaining the representative of CURES, was invited to participate and 761 participated (97.2% response rate). This included, 332 subjects without MS (non-MS group) and 422 subjects with MS (MS group). Subjects with infectious or inflammatory diseases and those on statins and asprins were excluded from the study.

Metabolic syndrome (MS) was defined according to the National Cholesterol Education Program–Adult Treatment Panel III criteria (Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults, 2001) modified for waist according to World Health Organization Asia Pacific guidelines for obesity as shown below (World Health Organization & International Association for the Study of Obesity and International Obesity Task Force, 2000). MS was defined as the presence of any three of the following abnormalities: abdominal obesity defined as waist circumference ≥90 cm for men and ≥80 cm for women, high blood pressure (systolic blood pressure ≥130 mmHg or diastolic blood pressure ≥85 mmHg and/or on antihypertensive medications), elevated fasting glucose (fasting plasma glucose ≥100 mg/dL and/or on anti-diabetic medications), hypertriglyceridemia (≥150 mg/dl), or low high-density lipoprotein-cholesterol (<40 mg/dL for males and <50 mg/dL for females).

Table 1
Clinical and biochemical characteristics of study subjects.

Parameters	Non-MS (n = 332)	MS (n = 422)	*p*
Age [years]	45.3 ± 14.4	50.6 ± 10.6	<0.001
Gender (Male) (%)	61.2	49.2	0.001
Waist circumference [cm]	84.0 ± 10.6	92.2 ± 9.4	<0.001
Body mass index [kg/m^2]	23.3 ± 4.1	26.3 ± 4.0	<0.001
Systolic blood pressure (SBP) [mmHg]	117.7 ± 16.5	133.1 ± 12.1	<0.001
Diastolic blood pressure (DBP) [mmHg]	73.2 ± 10.3	79.0 ± 11.7	<0.001
Fasting plasma glucose [mg/dl]	111.9 ± 61.1	156.4 ± 66.7	<0.001
Glycated hemoglobin (HbA1c) [%]	6.8 ± 2.4	8.4 ± 2.3	<0.001
Total cholesterol [mg/dl]	100.1	175.5	<0.001
Serum triglycerides [mg/dl]*	110.9 ± 64.8	204.5 ± 147.0	<0.001
High density lipoprotein [HDL] cholesterol [mg/dl]	44.8 ± 10.7	39.2 ± 7.9	<0.001
Low density lipoprotein [LDL] cholesterol [mg/dl]	119.0 ± 36.5	122.4 ± 41.1	0.237
Microalbuminuria [mg/dl]	20.3 ± 38.7	36.4 ± 54.0	<0.001

* Values are expressed as geometric mean.

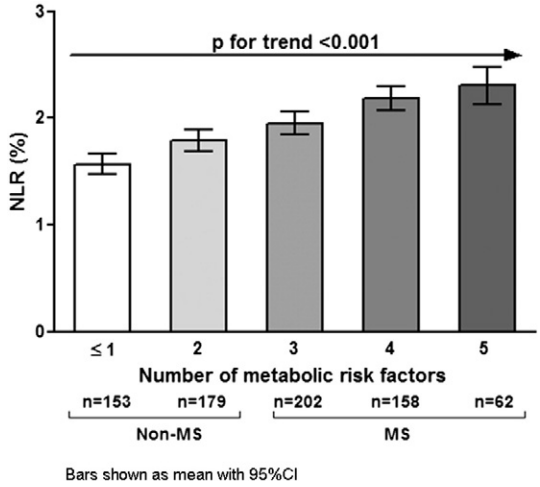

Fig. 1. NLR levels in relation to number of metabolic abnormalities.

Institutional ethical committee approval was obtained for the study, and informed consent was obtained from all the study subjects.

2.1. Anthropometric measurements

Anthropometric measurements, i.e. height, weight, and waist, were obtained using standardized techniques as detailed elsewhere (Deepa et al., 2003). Height was measured with a tape measured to the nearest centimeter. Weight was measured with a traditional spring balance that was kept on a firm horizontal surface. Waist was measured using a nonstretchable fiber measuring tape. The body mass index (BMI) was calculated as the weight (in kg) divided by the square of the height (in m). Blood pressure was recorded in the right arm in the sitting position to the nearest 2 mmHg with a mercury sphygmomanometer (Diamond Deluxe BP apparatus, Industrial Electronic and Allied Products, Pune, India). Two readings were taken 5 min apart, and the mean of the two was taken as the blood pressure.

2.2. Biochemical parameters

Fasting plasma glucose (glucose oxidase–peroxidase method), serum cholesterol (cholesterol oxidase–peroxidase-amidopyrine-method), serum triglycerides (glycerol phosphate oxidase–peroxidase-amidopyrine-method), high-density lipoprotein-cholesterol (direct method; polyethylene glycol–pretreated enzymes), and creatinine (Jaffe's method) were measured using a Hitachi-912 Autoanalyzer (Boehringer Mannheim/Hitachi, Mannheim, Germany). The intra- and interassay coefficients of variation for the biochemical assays ranged between 3.1% and 7.6%. Low-density lipoprotein cholesterol was calculated using the Friedewald formula. Glycated hemoglobin (HbA1c) was estimated by high pressure liquid chromatography using a Variant machine (Bio-Rad, Hercules, CA). The intra- and interassay coefficients of variation of HbA1c were less than 10%.

2.3. Measurement of leukocyte count and NLR

Leukocyte count was assessed using a five-part hematology analyzer (model SF3000; Sysmex, Kobe, Japan) based on flow cytometry. The intra- and interassay coefficients of variation of the leukocyte count were <10%. NLR was calculated as the ratio between (i.e. percentage of) neutrophils and total lymphocyte counts in the study subjects.

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